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Veterinary Parasitology 112 (2003) 261–267

veterinary
parasitology

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Clinical *Sarcocystis neurona* encephalomyelitis in a domestic cat following routine surgery

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Received 25 October 2002; received in revised form 9 January 2003; accepted 10 January 2003

Abstract

Sarcocystis neurona is an important cause of equine protozoal myeloencephalitis (EPM) in horses in the Americas. An EPM-like neurological disease also has been reported from other mammals but it is difficult to induce this disease in the laboratory. A 4-month-old male domestic cat developed neurological signs 3 days following castration. The cat was euthanized 12 days later because of paralysis. Encephalomyelitis was the only lesion and was associated with numerous *Sarcocystis* schizonts and merozoites in the brain and spinal cord. The protozoa reacted positively with *S. neurona*-specific polyclonal rabbit antibody. Two unidentified sarcocysts were present in the cerebellum. It may be possible that stress of surgery triggered relapse of *S. neurona* infection in this cat. © 2003 Elsevier Science B.V. All rights reserved.

Keywords: *Sarcocystis neurona*; Neurological; Encephalomyelitis; Schizonts; Sarcocysts; Cat; *Felis domesticus*

1. Introduction

Sarcocystis neurona is the most important cause of a serious neurological disease of horses, called equine protozoal myeloencephalitis (Dubey et al., 1991, 2001b). It also causes EPM-like disease in other mammals including mink, raccoons, cats, skunks, sea otters and Pacific harbor seals (Dubey et al., 2001b). The life cycle of *S. neurona* is not fully known. Opossums (*Didelphis virginiana*, *Didelphis albiventris*) are its definitive hosts. Horses are considered its aberrant hosts because only schizonts and merozoites (no sarcocysts) are

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found in horses. Recently, domestic cats, sea otters, armadillos, skunks, and raccoons were shown to be intermediate hosts for *S. neurona* (Dubey et al., 2001b). Experimentally, it has been difficult to induce clinical *S. neurona* infection in any animal other than immunodeficient mice (for review see Dubey et al., 2001b). We report clinical encephalomyelitis in a cat following a routine surgical operation that is likely due to *S. neurona*.

2. Materials and methods

A 4-month-old mixed breed male cat was brought to the Tender Care Animal Hospital (TCAH), Morton, Illinois for routine castration and onychectomy. The cat recovered completely from the anesthesia and was sent home. The cat was noticed to have neurological signs on the 3rd day after surgery and was walking sideways when brought to the TCAH on the 5th day after surgery. The cat was euthanized on the 12th day after surgery because of posterior paralysis.

A necropsy was performed on the cat at the Animal Disease Laboratory, Galesburg, 24 h after euthanasia. Specimens of heart, lung, spleen, liver, intestine, kidney, mesenteric lymph nodes, spinal cord, and brain were fixed in 10% buffered neutral formalin. Paraffin-embedded sections were cut at 5 μm thickness, stained with hematoxylin and eosin, and examined microscopically. Retrospectively, paraffin sections were examined by immunohistochemistry using rabbit polyclonal antibodies against *Toxoplasma gondii*, *Neospora caninum* and *Sarcocystis neurona* with methods described previously (Lindsay and Dubey, 1989; Dubey and Hamir, 2000). Positive and negative controls were included for each organism. Preparation of anti-*S. neurona* antibody and its specificity were described by Dubey and Hamir (2000). Although DNA analysis is now the standard for distinguishing *S. neurona* from other *Sarcocystis* sp., unfixed tissues were not available for DNA extraction.

3. Results

Gross lesions were not seen. Microscopically, lesions were confined to the brain and spinal cord and were more severe in the latter (Figs. 1 and 2). In brain stem and cerebrum there were multiple foci characterized by mild to heavy perivascular cuffs of lymphocytes and a few macrophages with gliosis and variable spongiosis of the adjacent neuropil. Similar lesions were present in cervical, lumbar, and thoracic spinal cord with severe involvement of the meninges; perivascular cuffs and gliosis involving both gray and white matter (Figs. 1 and 2).

Sarcocystis stages (sarcocysts, schizonts) were seen in the brain and spinal cord. Two sarcocysts were seen in one section of cerebellum (Fig. 3). These sarcocysts were 90 μm \times 80 μm and 170 μm \times 80 μm in size, and were located in cortex without any host reaction. The sarcocyst wall was approximately 2 μm thick and had villar protrusions (Fig. 3). Sarcocysts were mature and contained slender 5 μm \times 1 μm bradyzoites. No other details were available because sarcocysts were not seen in any of the subsequent sections.

Schizonts were located in and around neural lesions. Most schizonts were mature and contained merozoites that were arranged peripherally in a rosette or more commonly haphazardly (Fig. 1). Immature schizonts contained a central large nucleus with a prominent

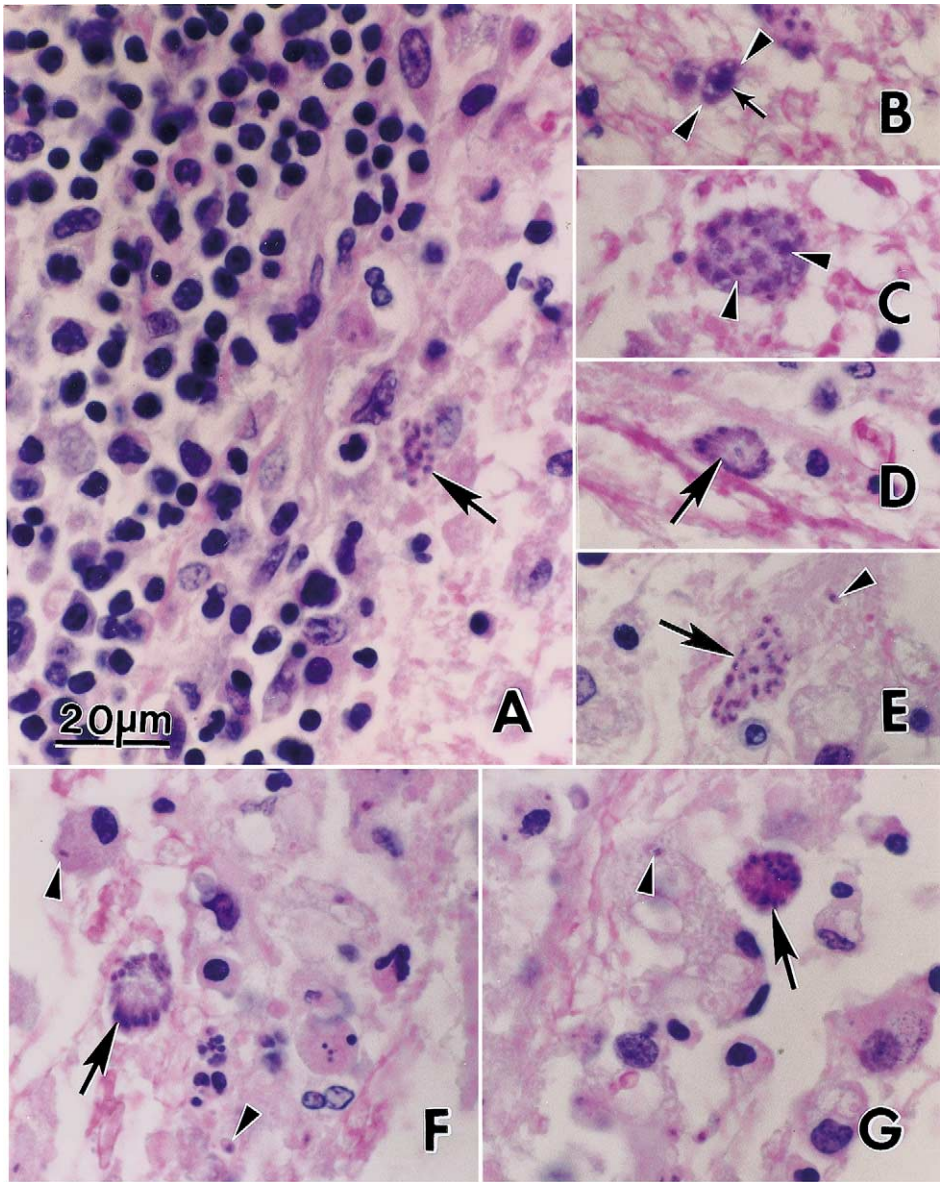


Fig. 1. *Sarcocystis neurona* in sections of spinal cord of the naturally-infected cat (H and E). (A) Mononuclear cell infiltration in meninges. Arrow points to a schizont; (B) uninucleate organism (arrowheads) with a large nucleus and a nucleolus (arrow); (C) schizont with several nucleoli (arrowheads); (D) schizont merozoites at the periphery; (E) mature schizont (arrow) and a free merozoite (arrowhead); and (F, G) schizonts (arrows) and individual merozoites (arrowheads) among necrotic host cells.

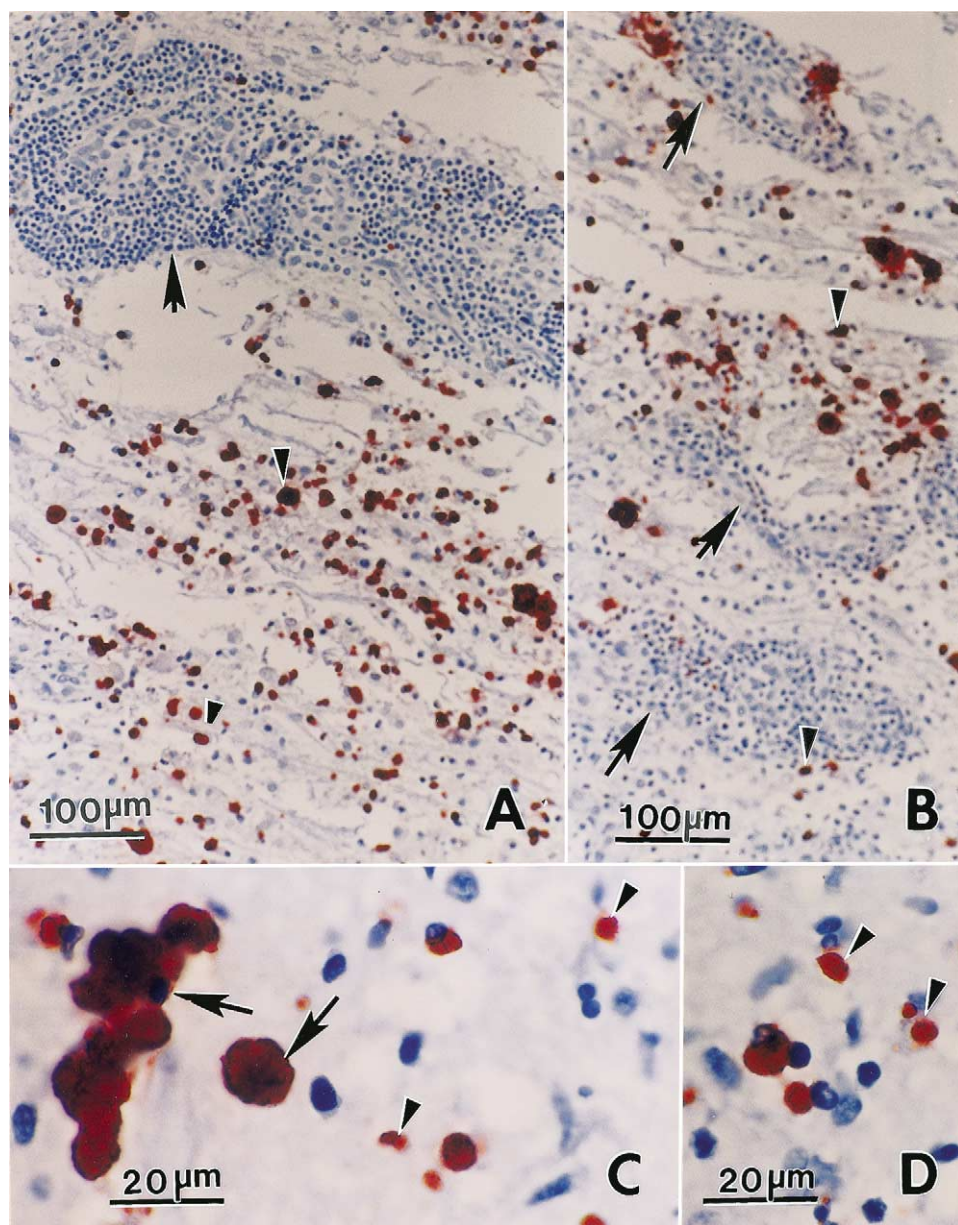


Fig. 2. *Sarcocystis neurona* (all red spots) in sections of spinal cord of the naturally-infected cat. Immunohistochemical stain with anti-*S. neurona* antibody. (A, B) Severe perivascular infiltration (arrows) and numerous organisms (arrowheads) and (C, D) schizonts (arrows) and individual merozoites (arrowheads).

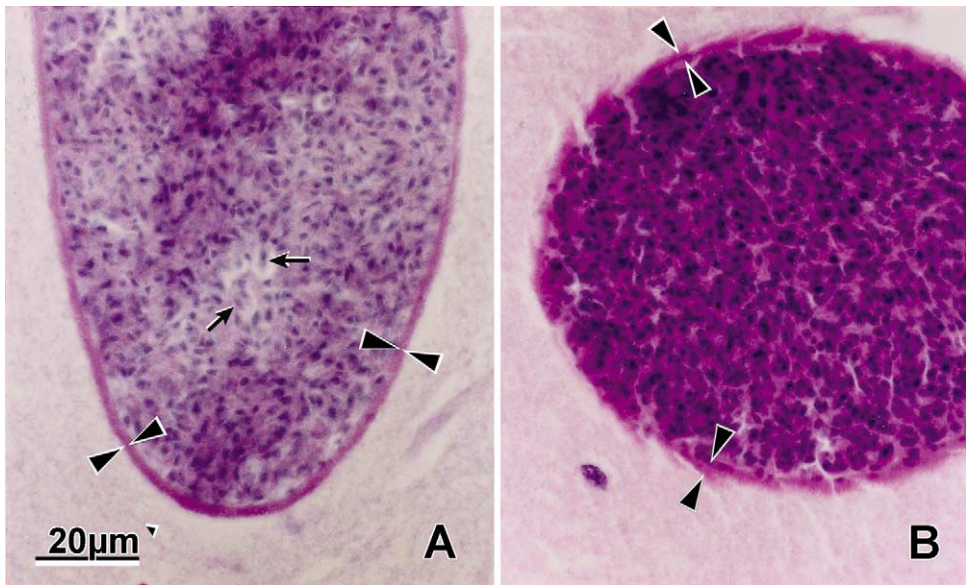


Fig. 3. *Sarcocystis* sp. sarcocysts in a section of the cerebellum of the naturally-infected cat (H and E). Note thick sarcocyst cyst wall (arrowheads) enclosing slender, small bradyzoites (arrows).

nucleolus and one host cell contained several immature schizonts (Fig. 1C). There were free merozoites scattered in and around lesions. Schizonts and merozoites were present in neural cells, mononuclear cells and few merozoites were seen in mononuclear cells within the meninges.

Numerous schizonts and merozoites were seen in sections stained with anti-*S. neurona* antibody (Fig. 2) compared with sections stained with H and E. Organisms were not stained with antibodies to *T. gondii* and *N. caninum*.

Serologic tests for antibodies to feline leukemia virus, feline immunodeficiency virus, and feline infectious peritonitis virus were negative and there was no evidence for immunodeficiency syndrome.

4. Discussion

Schizonts and merozoites in lesions of brain and spinal cord were identified as *S. neurona* based on morphology of the organisms, tissues parasitized, and reactivity to *S. neurona*-specific antibody although DNA analysis was lacking. *Sarcocystis neurona* schizonts in horses and other naturally infected animals have been found generally in the central nervous system (CNS) as was the case in this cat. The parasite divided by endopolygony, a process where the nucleus becomes multi-lobed and gives rise to more than four merozoites whereas in *T. gondii* and *N. caninum* the nucleus divides and forms two organisms. Additionally, the protozoa did not react to *T. gondii* and *N. caninum* antibodies. The

identification of the parasite was essentially based on immunoreactivity to *S. neurona* polyclonal antibody that is not known to react with any related protozoa (Dubey and Hamir, 2000). Unfixed tissues were not available for DNA analysis.

The onset of neurological signs in the present cat, 3 days following surgery, is of interest because apparently normal horses have fallen ill to *S. neurona* while racing (Daft et al., 2002) and stress can aggravate clinical signs in EPM-affected horses (Saville et al., 2001). There was no evidence of any immune suppression before surgery but owners declined to perform any hematological evaluation before surgery.

Sarcocystis neurona encephalitis was reported previously in a 3-month-old cat (*Felis domesticus*) from California (Dubey et al., 1994). The cat became lame after a fall and developed hemiparesis. Protozoa were confined to the brain and spinal cord. Neurological *S. neurona* infection has also been reported in a 13-year-old captive Canadian lynx (*Felis lynx Canadensis*) (Forest et al., 2001).

Two sarcocysts found in the cerebellum of the cat in the present study could not be identified further. The sarcocysts appeared to be structurally similar to *S. neurona* sarcocysts in experimentally-infected cats (Dubey et al., 2000, 2001a). Sarcocysts were not available for ultrastructural or immunological investigations. Unfortunately, tongue and other skeletal muscles from this cat were discarded before histological evaluation.

Serologic and parasitological studies indicate that the cat can be involved in the life cycle of *S. neurona*. Laboratory-raised cats fed *S. neurona* sporocysts developed patent *S. neurona* infection; schizonts were detected in histological sections of the cat and sarcocysts matured in their muscles (Dubey et al., 2000). Laboratory-reared opossums shed *S. neurona* sporocysts after feeding muscle tissue from experimentally-infected cats (Dubey et al., 2000). Turay et al. (2002) found *S. neurona* sarcocysts in a naturally infected cat that were infective to a laboratory-reared opossum. The sporocysts shed by the opossum were identified as *S. neurona* based on bioassay in immune deficient mice. The strain of *S. neurona* isolated from the cat was infective to cats (Butcher et al., 2002). Rossano et al. (2002) reported antibodies to *S. neurona* in 5% of 196 domestic cats from Michigan whose sera were submitted for *T. gondii* antibody analysis, and Stanek et al. (2003) found *S. neurona* antibodies in 13% of 310 farm cats from Ohio. Thus, there is evidence of natural exposure of *S. neurona* infection in cats. Results of the present study indicate that *S. neurona* should be considered in differential diagnosis of neurological diseases in cats, and antiprotozoal therapy may be appropriate as it is known to alleviate suffering due to EPM in horses (Dubey et al., 2001b).

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